CLAIMS:

1. Use of an Id gene product in promoting self-renewal of pluripotent cells in culture.

2. Use according to Claim 1 of a combination of the Id gene product with an activator of a gp130 downstream signalling pathway.

3. Use of a combination of
   (i) an agent that increases Id protein expression or activity; and
   (ii) an activator of a gp130 downstream signalling pathway,
   in promoting self-renewal of pluripotent cells in culture.

4. Use according to any of Claims 1-3, wherein the activator of a gp130 downstream signalling pathway is LIF.

5. Use according to any of Claims 1-4, wherein the pluripotent cells are embryonic stem cells.

6. Use according to Claim 5 wherein the embryonic stem cells are mouse cells or human cells.

7. Use according to any of Claims 3-6 wherein the agent (i) is selected from fibronectin, agonists of the fibronectin receptor, activators of integrin signalling, nanog, and homologes of all of the aforementioned that induce Id gene expression or Id protein activity.

8. Use according to any of Claims 1-7, comprising inducing expression of an Id gene.

9. Use according to any of Claims 1-8, comprising genetically manipulating a pluripotent cell so that it expresses an Id gene.

10. Use according to any of Claims 1-9, comprising introducing into a pluripotent cell a vector comprising an Id gene.

11. Use according to any of Claims 1-11 wherein the Id gene product is an Id protein.

12. A method of promoting self-renewal of a pluripotent cell in culture, comprising (1) expressing an Id gene or inducing expression of an Id gene in the cell, or culturing the cell in medium containing an Id protein, and (2) activating GP130 downstream signalling.

13. A method according to Claim 12, comprising expressing an Id gene episomally in the cell.

14. A method according to Claim 13 comprising expressing an Id gene from an episomal vector comprising an inducible promoter.

15. A method according to any of Claims 12-14, comprising stimulating gp130 downstream signalling by culturing the cell in medium comprising a cytokine acting through gp130.

16. A method according to Claim 15 wherein the cytokine is selected from LIF, CNTF, Cardiotrophin, Oncostatin M and a combination of IL-6 plus sIL-6 receptor.

17. Use of a combination of:-

(a) a direct activator or effector of Id gene expression and/or Id protein activity, other than one acting through a receptor of the TGF-β superfamily; and

(b) an activator of a gp130 downstream signalling pathway, in promoting self-renewal of a pluripotent cell in culture.

18. A method of culture of ES cells so as to promote ES cell self renewal, comprising maintaining the ES cells in medium containing:-
(a) an Id protein or a direct activator or effector of Id gene expression and/or Id protein activity, other than one acting through a receptor of the TGF-β superfamily; and
(b) an activator of a gp130 downstream signalling pathway.

19. A method of culture of ES cells, comprising:-
(a) maintaining the ES cells in a pluripotent state in culture, optionally on feeders, in the presence of a cytokine acting though gp130 and serum or an extract of serum;

(b) passaging the ES cells at least once;

(c) withdrawing the serum or the serum extract from the medium and withdrawing the feeders if present, so that the medium is free of feeders, serum and serum extract; and

(d) subsequently maintaining ES cells in a pluripotent state in the presence of:-
(i) a direct activator or effector of Id gene expression and/or Id protein activity, other than one acting through the receptor of the TGF-β superfamily; and
(ii) an activator of a gp130 downstream signalling pathway.

20. A method of obtaining a transfected population of ES cells, comprising:-
(a) transfecting ES cells with a construct encoding a selectable marker operably linked to a promoter that expresses the selectable marker preferentially in ES cell;

(b) plating the ES cells;

(c) culturing the ES cells in the presence of

(i) a direct activator or effector of Id gene expression and/or Id protein activity, other than one activator acting through a receptor of the TGF-β superfamily; and

(ii) an activator of a gp130 downstream signalling pathway; and

(d) selecting for cells that express the selectable marker.

21. A method of culture of ES cells, comprising transferring an individual ES cell to a culture vessel and culturing the ES cell in the presence of

(a) a direct activator or effector of Id gene expression and/or Id protein activity, other than one acting through a receptor of the TGF-β superfamily; and

(b) an activator of a gp130 downstream signalling pathway,

so as to obtain a clonal population of ES cells, all of which are progeny of a single ES cell.

22. A method of directing differentiation of an ES cell towards a non-neurectodermal fate, comprising:-

(a) maintaining the ES cell in the presence of a cytokine acting through gp130 and a direct activator or effector of Id gene expression and/or Id protein activity, other than one acting through a receptor of the TGF-β superfamily; and

(b) withdrawing the cytokine whilst

(c1) maintaining the direct activator or effector of Id gene expression and/or Id protein activity; and/or

(c2) adding a further signalling molecule capable of directing differentiation.

23. A medium for self-renewal of ES cells, comprising:-

(1) basal medium;

(2) a direct activator or effector of Id gene expression and/or Id protein activity, other than one acting through a receptor of the TGF-β superfamily;

(3) an activator of gp130 downstream signalling pathways; and

(4) an iron transporter;

wherein the medium is free of serum or serum extract.

24. A method of deriving a pluripotent cell from a blastocyst, comprising:-

(1) obtaining a blastocyst;

(2) culturing the blastocyst in the presence of an activator of gp130 downstream signalling, to obtain an inner cell mass;

(3) dissociating the inner cell mass;

(4) isolating a cell from the dissociated inner cell mass; and

(5) culturing the isolated cell in the presence of an activator of gp130 downstream signalling and an activator of Id gene expression or a product of Id gene expression.

25. A method according to Claim 24, comprising culturing the blastocyst in LIF.

26. A method according to Claim 24 or 25 comprising culturing the isolated cell in a combination of LIF and an agonist of the BMP receptor.

27. A method according to any of Claims 24-26, comprising culturing the blastocyst for a period of from 2 to 4 days.

28. A method according to any of Claims 24-27 comprising culturing the isolated cell in serum free medium.

29. A method according to any of Claims 24-28 comprising culturing the blastocyst in serum free medium.

30. A method according to any of Claims 24-29 comprising culturing the blastocyst in the absence of an agonist of the BMP receptor.

31. A vector, comprising an Id gene operatively linked to a promoter.

32. A vector according to Claim 31 wherein the promoter is an inducible promoter.

33. A vector according to Claim 31 or 32 which is an episomal vector.

34. A culture medium comprising an agent which induces Id protein expression, other than an agent acting through a receptor of the TGF-β superfamily of receptors.

35. A culture medium comprising an Id protein.

36. A culture medium according to Claim 35, comprising an Id protein linked to a translocation domain, to facilitate translocation of the Id protein across the cell membrane of a pluripotent cell.

37. A culture medium according to claim 35 or 36, comprising an Id protein linked to TAT, VP22 or a penetratin.

38. A composition, comprising an Id protein and a translocation domain.

39. A composition according to claim 38, comprising a fusion protein.

40. A composition according to claim 38 or 39, wherein the translocation domain comprises TAT, VP22 or a penetratin.

41. Use of an agent that increases Id protein activity in a pluripotent cell, in promoting self-renewal of the pluripotent cell.

42. Use according to Claim 41 wherein the agent increases the amount of Id protein in the cell.

43. Use according to Claim 41 wherein the agent comprises a composition according to any of claims 38 to 40.

44. A cell obtained by *in vitro* culture of a pluripotent cell in the presence of gp130 signaling and activation and/or expression of an Id protein.

45. A method of obtaining a differentiated cell, comprising
(1a) expressing an Id gene or inducing expression of an Id gene in a cell, and
(1b) activating GP130 downstream signalling in the cell;
(2) differentiating the cell; and
(3) obtaining a differentiated cell.

46. A method according to claim 45, wherein the cell of step (1) comprises a construct in which a nucleotide sequence encoding a selectable marker is operatively linked to a promoter which preferentially expresses the selectable marker in a desired cell.

47. A method according to claim 46, comprising selecting for cells expressing the selectable marker.

48. A cell obtained by a method according to any of claims 45 to 47.

49. A method of obtaining a pluripotent cell, comprising

expressing an Id gene or inducing expression of an Id gene in a cell, or culturing a cell in medium containing an Id protein, and activating gp130 downstream signalling in the cell, wherein the cell is obtained from somatic cells or tissue of a fetus or adult.

50. A method according to claim 49, wherein the pluripotent cell is characterised by being positive for Rex1, Oct4 and nanog.

51. A cell obtained by a method according to any of claims 49 to 50.

52. An assay for a factor with activity that substitutes for an Id protein, comprising:-

(1) culturing a cell in the presence of Id protein activity and gp130 downstream signaling, thereby maintaining the cell in a pluripotent state;

(2) removing or reducing the Id protein activity;

(3) introducing the factor to the cell; and

(4) determining whether the cell remains pluripotent or differentiates.

53. An assay according to Claim 52, wherein culturing the cell in the presence of Id protein activity in (1) comprises (a) expressing an Id gene, (b) inducing expression of an Id gene or (b) adding an Id protein to medium in which the cell is cultured.

54. An assay according to claim 52 or 53, wherein introducing the factor to the cell comprises (a) expressing the factor, or (b) adding the factor to medium in which the cell is cultured.

55. A factor obtained by the method of any of claims 52 to 54.